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Per- and polyfluoroalkyl substances (PFAS) exposure in plasma and their blood–brain barrier transmission efficiency–A pilot study

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ABSTRACT

Per- and polyfluoroalkyl substances (PFAS) have been shown to penetrate the blood–brain barrier (BBB) and accumulate in human brain. The BBB transmission and accumulation efficiency of PFAS, as well as the potential health risks from human co-exposure to legacy and emerging PFAS due to differences in transport efficiency, need to be further elucidated. In the present pilot study, 23 plasma samples from glioma patients were analyzed for 17 PFAS. The concentrations of PFAS in six paired brain tissue and plasma samples were used to calculate the BBB transmission efficiency of PFAS (R_{PFAS}). This R_{PFAS} analysis was conducted with utmost care and consideration amid the limited availability of valuable paired samples. The results indicated that low molecular weight PFAS, including short-chain and emerging PFAS, may have a greater potential for accumulation in brain tissue than long-chain PFAS. As an alternative to perfluorooctane sulfonic acid (PFOS), 6:2 chlorinated polyfluorinated ether sulfonate (6:2 Cl-PFESA) exhibited brain accumulation potential similar to that of PFOS, suggesting it may not be a suitable substitute concerning health risk in brain. The BBB transmission efficiencies of perfluorooctanoic acid, PFOS, and 6:2 Cl-PFESA showed similar trends with age, which may be an important factor influencing the entry of exogenous compounds into the brain. A favorable link between perfluorooctane sulfonamide (FOSA) and the development and/or progression of glioma may be implicated by a strong positive correlation ($r^2 = 0.94$; $p < 0.01$) between R_{FOSA} and Ki-67 (a molecular marker of glioma). However, a causal relationship between R_{FOSA} and glioma incidence were not established in the present study. The present pilot study conducted the first examination of BBB transmission efficiency of PFAS from plasma to brain tissue and highlighted the importance of reducing and/or controlling exposure to PFAS.

1. Introduction

Per- and polyfluoroalkyl substances (PFAS) are synthetic chemicals that have been extensively used in a broad variety of processes and products, such as electroplating, textile, and printing and dyeing industries (Liu et al., 2022; Liu et al., 2021b; Wang et al., 2023), for over 70 years. An increasing number of PFAS have been found to possess persistency, bioaccumulative ability, and toxicity (PBT) (Jiao et al., 2023; Liu et al., 2021b); for example, 21.8 % of PFAS from a database of emerging PFAS obtained from machine learning models were found to exhibit PBT attributes (Han et al., 2023). Consequently, perfluorooctane sulfonic acid (PFOS), perfluorooctanoic acid (PFOA), and

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perfluorohexane sulfonic acid (PFHxS) were placed on the regulation list by the Stockholm Convention in 2009, 2019, and 2021, respectively (Friederichs et al., 1996; UNEP, 2009; UNEP, 2019; UNEP, 2022). Although PFOA and PFOS have been regulated for a number of years, they remain frequently detected in the global environment and humans. For example, perfluorooctane sulfonamide (FOSA) has been found in over 80 % of wild freshwater fishes in the Pearl River Delta of China (Pan et al., 2014) and can degrade into perfluorobutane sulfonic acid (PFBS), PFHxS, and PFOS (Zhao et al., 2018). The substitutes of PFAS, such as 6:2 chlorinated polyfluorinated ether sulfonate (6:2 Cl-PFESA), perfluoro-2-propoxypropanoic acid, and ammonium 4,8-dioxa-3H-perfluorononanoate, have also been recognized as emerging contaminants because their exposure levels and health risks are comparable to those of traditional PFAS (Li et al., 2018; Liu et al., 2021a). These findings highlight the need to further monitor the potential risks of PFAS and to develop appropriate management and control strategies.

Per- and polyfluoroalkyl substances are found to pose toxic risks to various human organs, including but not limited to thyroid, liver, kidney, and brain (Fenton et al., 2021; Xie et al., 2023). Notably, brain, being a vital and highly protected organ, is particularly vulnerable to these exogenous substances (Abbott et al., 2010). Despite the presence of the blood–brain barrier (BBB), PFAS can penetrate the BBB and accumulate in both infant and adult brains (Liu et al., 2021b; Maestri et al., 2006; Pérez et al., 2013; Xie et al., 2022). Per- and polyfluoroalkyl substances can enter the brain through substance exchange in the cerebral circulation (Cao et al., 2021) and/or transporter-mediated active transport such as binding to transporters (Abbott et al., 2006; Cao et al., 2021). An incomplete or damaged BBB may allow exogenous substances to invade the brain, as seen in developing brain barriers in fetus (Liu et al., 2021b), and compromise brain barriers in brain disease patients (Rosenberg, 2012). However, our current understanding of the BBB transmission efficiency of PFAS remains limited. Only a few studies have explored the BBB transmission of PFAS by comparing the concentrations of PFAS in human blood and cerebrospinal fluid (CSF) samples (Liu et al., 2021b; Shang et al., 2023; Wang et al., 2018). A recent study utilizing autopsy samples in Denmark provided data on the concentration ratios of four PFAS (PFOA, perfluorononanoic acid (PFNA), PFHxS, and PFOS) in human organs (including brain) and whole blood (Nielsen et al., 2024).

Our previous studies investigated the levels of PFAS exposure and their potential association with brain tissue pathologies (Xie et al., 2022; Xie et al., 2023). To fill the above-mentioned knowledge, we provided the first illustration of the BBB transmission efficiency based on the concentration ratios of PFAS in paired brain tissue and plasma samples. We delved into the underlying factors that motivated our previous studies, specifically examining the permeability of the BBB and the ability of PFAS to accumulate in the brain. Furthermore, we examined any potential correlation between the BBB transmission efficiency of PFAS and the risk of glioma tumorigenesis. The findings from these analyses on PFAS transmission efficiency and its correlation with glioma tumorigenesis risk should provide valuable insights that can contribute to the development of effective treatment strategies for individuals diagnosed with brain tumors.

2. Materials and methods

2.1. Materials

Ten target perfluoroalkyl carboxylic acids (PFCAs; Table S1) with 4 − 13 carbons were acquired from Wellington Laboratories (Guelph, Ontario, Canada). Two perfluorinated sulfonic acids, PFBS (C4) and PFHxS (C6), were obtained from Toronto Research Chemicals (Toronto, Ontario, Canada). The PFOS (C8) standard solution was purchased from AccuStandard (New Haven, CT, USA). Perfluorooctane sulfonamide and three emerging PFAS, including perfluoro-2-propoxypropanoic acid, 6:2 Cl-PFESA, and 8:2 chlorinated polyfluorinated ether sulfonate, were provided by Wellington Laboratories. Surrogate standards made up of seven carbon-labeled standards (Table S1) and perfluoro-n-[$^{13}C_8$] octanoic acid and sodium perfluoro-1-[$^{13}C_8$] octanesulfonate used as internal standards were also purchased from Wellington Laboratories. More data on all target compounds, that is, molecular formula, CAS number, and octanol–water partition coefficient (K_{ow}), are summarized in Table S1. Commercial standard serum was purchased from Kangweishi Medical Devices (Shijiazhuang, Hebei, China) to examine matrix effects on pretreatment methods.

2.2. Sample collection and preparation

Twenty-three glioma patients from the Southern Hospital of Southern Medical University (Guangzhou, China) were recruited from 2019 to 2020. Plasma was used as a favorable blood matrix for determining the internal levels of PFAS in glioma patients due to practical considerations. Twenty-three plasma samples from the patients before operations were obtained. Whole blood was collected in K2-ethylenediaminetetraacetic acid anticoagulant vacutainer tubes and allowed to clot (>1h). Plasma was obtained by centrifugation at 3500 rpm for 3 min. All plasma samples were collected in polypropylene cryogenic vials and stored in a biorefrigerator at − 80 °C until analysis. The influence of anticoagulants was negligible in the present study (Jin et al., 2016). The study's objectives and contents were thoroughly explained to all participants. Approval for the present study was obtained from the Ethics Committees of Jinan University (Guangzhou, China; JNUKY-2022–008) and Southern Medical University (Guangzhou, China; NFGC-001-MCP).

Our previous study evaluated the effectiveness of the pretreatment method for brain tissue samples (Xie et al., 2022). In the present study, the extraction method of PFAS in plasma is similar to that employed by a previous study (Xie et al., 2022). Briefly, a 0.5 mL aliquot of plasma was spiked with 5 ng surrogate standards, and was combined with 1 mL of 0.5 mol/L tetrabutylammonium hydrogen sulfate (purity ≥ 97 %; Meryer Technologies, Shanghai, China) and 2 mL of 0.25 mol/L $NaHCO_3/Na_2CO_3$ buffer solutions. The mixture was left to equilibrate overnight and then sonicated in 4 mL of methyl *tert*-butyl ether for 20 min. Supernatants from three rounds of sonication-based extraction were separated through centrifugation and combined. The combined supernatant was concentrated under a gentle stream of nitrogen and subsequently redissolved in 0.5 mL of methanol. Before instrumental analysis, the final extract was spiked with the internal standards.

2.3. Instrumental analysis

The concentrations of target PFAS were determined in the negative ion mode using a Shimadzu LCMS-8050 triple-quadrupole system (Tokyo, Japan). An Ascentis C18 column (5 cm × 2.1 mm, 3 μm particle size; Bellefonte, PA, USA) was utilized with a 2 mM ammonium acetate (A)/acetonitrile (B) mobile phase for chromatographic separation. The gradient elution procedure was programmed with the following conditions: Starting with an initial composition of 10 % acetonitrile for 0.01 min, which was gradually increased to 90 % over a period of 0.5 min and maintained for 1 min. The composition of acetonitrile was then reduced to 60 % in 0.5 min, followed by a further reduction to 20 % in 0.5 min, and finally decreased to 10 % in 0.5 min and held steady for 4 min.

2.4. Quality assurance and quality control

All glass and fluorine-containing tubes were replaced with polypropylene containers to reduce potential interferences due to glass adsorption (Pérez et al., 2013). Standard human serum samples were validated to be free of the target PFAS before being used as matrix spiking samples. Procedural blank, solvent-spiked, and matrix-spiked samples were processed with the actual samples. The recoveries of the surrogate standards for procedural blank, solvent-spiked, matrix-spiked,

and actual plasma samples were 80 – 100 %, 80 – 100 %, 80 – 100 %, and 70 – 110 %, respectively, with standard deviations all less than 20 %. Recoveries of target PFAS in solvent-spiked samples ranged from 60 % to 120 %, with standard deviations less than 25 %. For matrix-spiked samples, recoveries of target PFAS were 60 %–120 %, with standard deviations less than 15 %. Detailed recovery data are listed in Table S2.

2.5. Data analysis

All measured values were corrected by those obtained in procedure blanks from the same batch, but not corrected by the recoveries of the surrogate standards. The reporting limit (RL) for PFAS in plasma samples, calculated through the lowest calibration concentration (0.1 ng mL^{-1}) divided by the actual sample volume (0.5 mL) and corrected for the final extract volume (0.5 mL), was 0.1 ng mL^{-1}. Concentrations of PFAS lower than RL were treated as zero for total concentrations calculation, and substituted with 0.5RL for composition analysis. Concentrations of PFAS in six glioma tissue samples that were paired with those in glioma plasma extracted from venous blood of glioma patients were adopted from a previous study (Xie et al., 2023). These six paired data were used in the present pilot study for evaluating the transmission efficiency of PFAS across BBB.

The BBB transmission efficiency of PFAS (R_{PFAS}) is defined as the concentration ratio between brain tissue and plasma (C_{brain}/C_{plasma}). The density of human plasma is estimated to be 1.0 kg L^{-1} and used to calculate R_{PFAS}. Hence, the different units between brain tissue and plasma were unified, similar to the concentration ratios for blood and placenta (Zhang et al., 2013). The R_{PFAS} was calculated using only paired samples where the concentrations in both brain tissue and plasma were higher than their RLs. Permeability of PFAS across the BBB was analyzed for four abundantly detected compounds, including PFOA, PFOS, FOSA, and 6:2 Cl-PFESA, as data were available for these four PFAS compounds. Regression analysis between R_{PFAS} and medical indicators, including tumor grade (characteristic of malignancy degree in a tumor), Ki-67 (nuclear antigens involved with multiplying cells), and P53 protein, was performed by IBM SPSS Statistics 24.0 (Chicago, IL, USA).

3. Results and discussion

3.1. Occurrence of PFAS in plasma of glioma patients

Among the 17 target PFAS, perfluorohexanoic acid (PFHxA), PFOA, PFNA, perfluorodecanoic acid (PFDA), PFOS, FOSA, and 6:2 Cl-PFESA, were identified in more than 60 % of the plasma samples from glioma patients (Table S3 and Fig. 1). The detection frequencies of PFAS in plasma samples were similar to those in brain tissue of glioma patients (Xie et al., 2022; Xie et al., 2023). Wang et al. (2020) also reported high detection frequencies (>80 %) of PFOA, PFNA, PFDA, PFOS, FOSA, and 6:2 Cl-PFESA in cord plasma of Beijing residents in North China, but they did not detect PFHxA. Different results have been reported for the detection of PFHxA. The detection frequency of PFHxA in the present study reached 65 %, while PFHxA was not detected in cord plasma samples from North China (Wang et al., 2020), nor in adult plasma samples from Norway (Poothong et al., 2017). However, our results were consistent with those of a recent study on the general population in Quanzhou (Liu et al., 2023), a southeastern city in China, where the detection frequencies of PFHxA in both plasma and red blood cells were 100 %. In the southern coastal city of Guangzhou, China, the high detection rate of PFHxA in human plasma samples may be attributed to frequent human consumption of PFHxA-enriched seafood, such as swimming crab (Gulkowska et al., 2006; Zhang et al., 2019), as well as different production modes of PFAS among different countries. Production of PFAS is currently undergoing a shift toward short-chain alternatives in China (Zhang et al., 2020). A previous study showed that PFHxA was rarely incorporated into consumer products in Norway

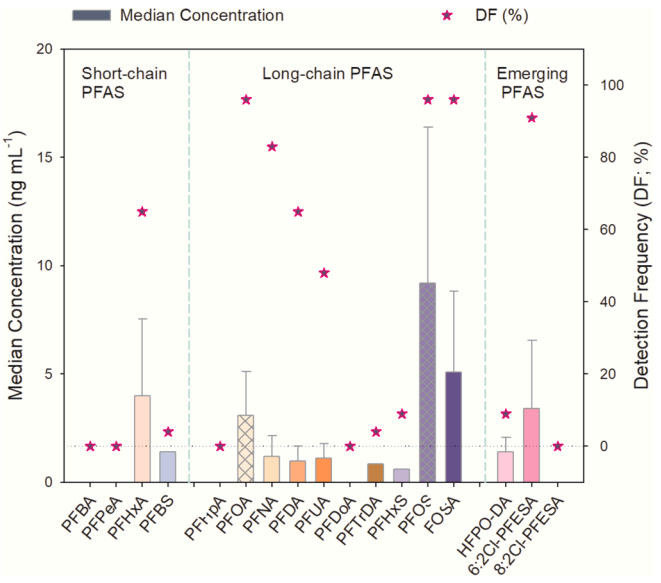

Fig. 1. Concentrations (ng mL^{-1}) and detection frequencies (DF; %) of perfluoroalkyl and polyfluoroalkyl substances (PFAS) in plasma samples from glioma patients. Notes: Full names for all PFAS are displayed in Table S1.

(Herzke et al., 2012).

The concentration of the sum of PFAS (∑PFAS) in plasma samples was 3.6 – 64 ng mL^{-1}, with a median of 28 ng mL^{-1} (Table S3). The sequence of median concentrations of PFAS was ∑PFSA (9.2 ng mL^{-1}) > ∑PFCA (8.9 ng mL^{-1}) > ∑Precursor (5.1 ng mL^{-1}) > ∑Emerging PFAS (3.6 ng mL^{-1}). This sequence was different from that of glioma brain tissue in our previous study, which was ∑PFCA > ∑PFSA > ∑Emerging PFAS > ∑Precursor (Xie et al., 2022). Concentration of PFHxA (4.0 ng mL^{-1}) as a short-chain PFCA (C < 7) was higher than those of PFBS (1.4 ng mL^{-1}) as a short-chain PFSA (C < 6) in plasma samples (Brendel et al., 2018). However, an opposite trend was observed for long-chain PFCAs and PFSAs. Similarly, the concentration of ∑Precursor was higher than that of ∑Emerging PFAS in plasma samples, but the order was reversed in glioma tissue (Xie et al., 2022). These results supported the notion that low molecular weight PFAS, including short-chain PFAS and emerging PFAS, could easily cross the BBB.

In the present study, PFOS exhibited the highest concentration (median: 9.2 ng mL^{-1}; mean: 10 ng mL^{-1}) in plasma, followed by FOSA (5.1 ng mL^{-1}; 5.4 ng mL^{-1}), PFHxA (4.0 ng mL^{-1}; 4.4 ng mL^{-1}), 6:2Cl-PFESA (3.4 ng mL^{-1}; 4.8 ng mL^{-1}), and PFOA (3.1 ng mL^{-1}; 3.3 ng mL^{-1}). Interestingly, PFOS concentration in the plasma of glioma patients was 3 – 6 times higher than those in cord plasma (mean: 1.5 ng mL^{-1}) from residents of northern China (Wang et al., 2020) and in plasma of the general population in southeastern China (mean: 2.6 ng mL^{-1}) (Liu et al., 2023). It is noteworthy that PFOS levels in glioma patients approached approximately half of the levels (age: 20 – 39; median: 18.1 ng mL^{-1}) found in the serum of residents near the DuPont Washington Works facility in the United States during the C8 Health Project (Frisbee et al., 2009). Furthermore, a case-control study in Chinese population (n = 1030) showed a positive association between PFOA and PFDA concentrations and incidences of breast cancer (Li et al., 2022), with PFOA and PFDA concentrations in cancer patients comparable to those observed in the present study. Our previous study (Xie et al., 2023) has evidenced that the accumulation of PFAS in brain tissue is associated with glioma. However, it remains to be confirmed if this association also extends to plasma, which is a fluid matrix.

A significant positive correlation ($r^2 = 0.95$; $p < 0.05$; Figure S1) was observed between PFOS concentrations in the paired brain tissue and plasma samples (Xie et al., 2023), suggesting that accumulation of PFOS in brain tissue occurs through the bloodstream. Therefore, PFOS can be

transported throughout the human body via blood, exerting systemic toxicity on multiple organs (Fenton et al., 2021), including the brain. For other compounds, significant associations between their concentrations in brain tissue and plasma could not be determined due to insufficient data. These findings underscore the significance of reducing and/or controlling exposure to PFAS in our daily life, as they have the potential to impact various organs, and highlight the need for further identifying the underlying mechanism governing the correlation.

3.2. Composition profiles of PFAS in paired brain tissue and plasma samples

Perfluorooctane sulfonic acid was the most abundant constituent among the 17 detected PFAS (Figure S2). The concentrations of PFOS, FOSA, PFOA, 6:2 Cl-PFESA, and PFHxA accounted for 40 %, 21 %, 13 %, 13 % and 5.4 % of the total concentration of PFAS, respectively, and the other 12 PFAS attributed less than 5 % of the total. Long-chain PFAS were the dominant class (Figure S2), accounting for 80 % of the total concentration. Emerging PFAS were identified as the second most abundant class (13 %), contributing to high exposure to 6:2 Cl-PFESA. The high exposure to 6:2 Cl-PFESA found in the present study can be attributed to the impacts of electroplating, textile, and dyeing industries in Guangdong. These industries have been flourishing in the region, and 6:2Cl-PFESA is constantly used as a substitute for PFOS in industrial processes (Liu et al., 2022).

The composition profiles of PFAS in both brain tissue and plasma (Xie et al., 2023) were dominated by long-chain PFAS when paired sample compositions are compared (Fig. 2). However, the abundance of low molecular weight PFAS was higher in brain tissue than in plasma. This result confirms the restricted effect of the BBB against macromolecular xenobiotics (Xie et al., 2022). The abundance of PFOA in plasma was found to be equivalent to that in paired brain tissue samples, whereas the abundances of PFOS and FOSA in plasma were twice as high as those in brain tissue. This difference in transmission between brain tissue and plasma suggests that PFCAs have a stronger transmission efficiency than PFSAs in penetrating the BBB, which is consistent with the placental transmission efficiency of PFAS (Gao et al., 2019).

3.3. Blood-brain barrier permeability of PFAS

The permeability of the BBB is low for endogenous metabolites and exogenous xenobiotics (Ge et al., 2005). The concentration ratios of PFAS between brain tissue and plasma can serve as an indicator to assess the transmission efficiency of exogenous substances (Wang et al., 2018). The calculated R_{PFAS} values for PFOA, PFOS, FOSA, and 6:2 Cl-PFESA were less than 0.4 (Table 1 and Fig. 3). This finding was consistent with those reported by previous studies (Harada et al., 2007; Maestri et al., 2006; Wang et al., 2018), indicating that the potential of PFAS penetrating the BBB is limited and accumulation in brain tissue is

Table 1
Information on participants who provided both glioma tissue and paired plasma samples and blood–brain barrier transmission efficiency of per- and polyfluoroalkyl substances (PFAS) (R_{PFAS} [a]).

Pairs	Age	Grade [b]	R_{PFOA}	R_{PFOS}	R_{FOSA}	$R_{6:2\ Cl\text{-}PFESA}$
1	17	IV	0.03	0.05	N.A. [c]	N.A.
2	27	IV	0.16	0.13	0.13	0.09
3	36	IV	0.08	0.07	0.17	0.07
4	48	IV	0.17	0.10	0.14	0.18
5	50	II	0.02	0.03	0.03	0.05
6	54	IV	0.37	0.06	0.09	0.28

Notes: Full names for all PFAS are displayed in Table S1.
[a] R_{PFAS} is defined as the concentration ratio between paired brain tissue and plasma samples.
[b] Glioma grade on a scale from I to IV, as per the classification criteria for central nervous system tumors established by the World Health Organization (Mao, 2016).
[c] N.A. = not available.

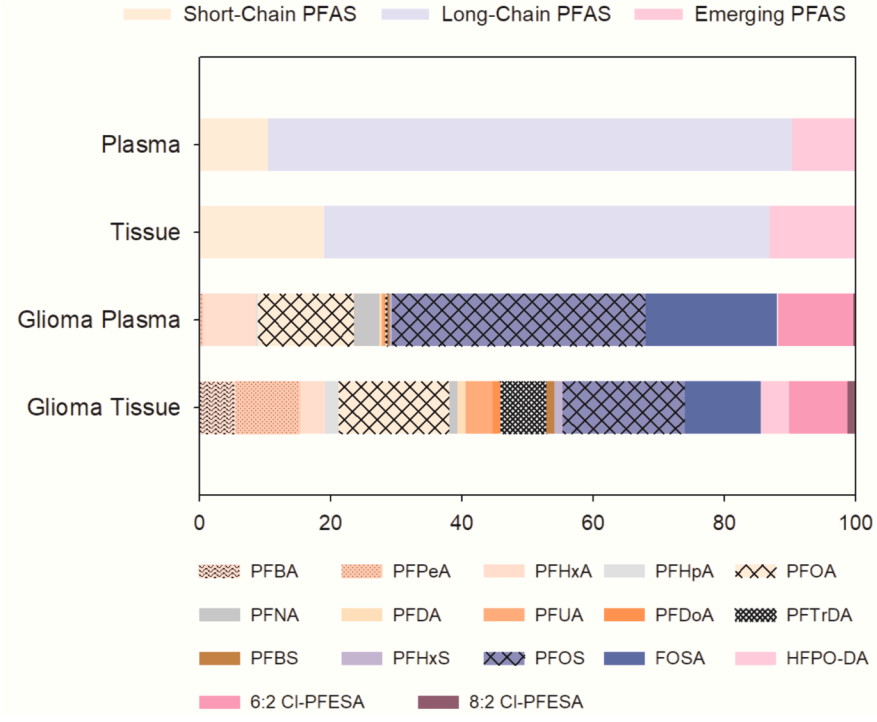

Fig. 2. Composition profiles (median; %) of PFAS in paired glioma plasma and glioma tissue samples (n = 6). Paired brain tissue data were). Notes: To enhance comparisons, different descriptions, such as plasma and glioma plasma, were utilized to differentiate between various combinations of PFAS. It is important to note that in this context, "plasma" is equivalent to "glioma plasma", and the total number of samples was six. The same principle applies to tissue and glioma tissue. adopted from (Xie et al., 2023

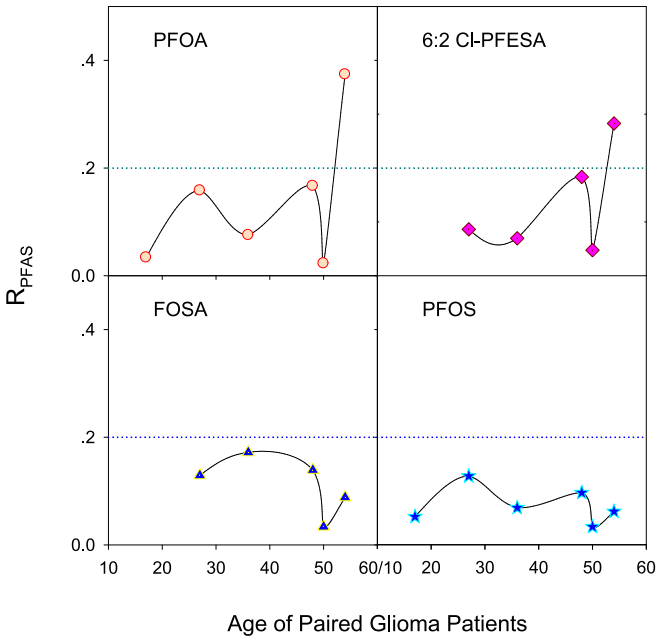

Fig. 3. Changes of blood–brain barrier transmission efficiency of PFAS (R_{PFAS}), defined as the concentration ratio in paired glioma tissue and plasma samples, with patients' ages.

insignificant. The R_{PFOA} (mean: 0.14; median: 0.12) in the present study was comparable to the concentration ratio of PFOA calculated in autopsy human brain tissue and blood (R_{PFOA}: 0.1667 in pooled) (Maestri et al., 2006). However, the R_{PFOA} and R_{PFOS} (mean: 0.07; median: 0.07) in the present study were much larger than the concentration ratios (C_{CSF}/C_{blood}) of PFAS in CSF and serum (Liu et al., 2021b; Wang et al., 2018). The mean values of R_{PFOA} and R_{PFOS} in nine pairs of neonatal CSF and serum samples (Liu et al., 2021b) were 0.0281 and 0.015, respectively, and were 0.0107 and 0.0121 in over 130 paired CSF and serum samples (Wang et al., 2018). The different results between C_{brain}/C_{blood} and C_{CSF}/C_{blood} could possibly be attributed to the long-term accumulation of PFAS in brain tissue rather than their short-term residence in CSF.

No significant differences were found between R_{PFOA} and R_{PFOS} in the present study (Friedman test; $p > 0.05$), similar to the result from a previous study where no variations in permeability were observed among PFOA, PFNA, PFDA, and perfluoroundecanoic acid (Wang et al., 2018). However, Liu et al., (2021b discovered that R_{PFOA} was higher than R_{PFOS} ($p < 0.05$) in newborns, because PFOS was tightly bound to serum albumin and therefore less likely to enter the brain (Beesoon and Martin, 2015). The BBB restricts the entry of PFOS, while other factors further limit the entry of PFOA, potentially resulting in no difference in the permeability of PFOA and PFOS.

Aside from the BBB, there are multiple reasons why exogenous compounds cannot penetrate the brain. Transport proteins and phospholipids have been identified as influencing factors that can alter the permeability of PFAS across the BBB (Beesoon and Martin, 2015; Dassuncao et al., 2019). Inflammation and barrier integrity are also crucial factors for the penetration of PFAS into the BBB (Wang et al., 2018). Given the presence of brain tumors, there is no guarantee that the brain barrier is completely unimpaired in patients with glioma (Arvanitis et al., 2020). Therefore, whether the BBB permeability of PFOA and PFOS is similar or different depends on the combined effects of multiple factors. The limited availability of human brain tissue and the inherent difficulty in obtaining paired brain-plasma samples underscore the unique and irreplaceable nature of these samples. As such, the present study was considered only a pilot one for assessing the PFAS transport efficiency in plasma-brain tissue. Further investigations are needed to gain better understanding of the BBB penetration mechanism of PFAS.

3.4. Age-related permeability of blood–brain barrier and connections between transmission efficiency and molecular markers

The BBB transmission efficiencies of PFOS and FOSA within the same participants were generally consistent with each other, with one exception where they varied by a large margin. The R_{PFOA}, R_{PFOS}, and $R_{6:2\ Cl\text{-}PFESA}$ in the present study diverged with age in a similar pattern (Fig. 3). For different participants, transmission efficiencies of PFOA, PFOS, and 6:2 Cl-PFESA exhibited similar trends with age, indicating that age could play a significant role in modulating the penetration of external substances into the brain. Probably because 6:2 Cl-PFESA has a small molecular weight and a strong affinity to albumin (Bischel et al., 2011; Sheng et al., 2020; Xie et al., 2022), the $R_{6:2\ Cl\text{-}PFESA}$ was equal to or higher than R_{PFOS}, which enables 6:2Cl-PFESA to transmit and accumulate in human brain more easily than PFOS. Previous studies suggested that 6:2 Cl-PFESA has greater bioaccumulation potential (Sheng et al., 2020), stronger sorption capacity by soils (Chen et al., 2018), and higher health risks than PFOS (Li et al., 2018; Lv and Sun, 2021). Therefore, 6:2 Cl-PFESA may be more harmful to brain than PFOS and should not be used as a substitute for PFOS.

A significant positive correlation ($r^2 = 0.94$; $p < 0.01$) between R_{FOSA} and Ki-67 was validated by a linear regression analysis (Fig. 4). In addition, the concentration differences of FOSA in brain tissue of different tumor grades were demonstrated in our previous study (Xie et al., 2023). Therefore, the BBB transmission efficiency of FOSA may be associated with the occurrence or development of glioma, as tumor grade and Ki-67 are trustworthy auxiliary indicators for the diagnosis of glioma (Arshad et al., 2010; Takano et al., 2016). No significant associations between PFAS and tumor or P53 were found in the present study. The accumulation of four perfluorinated substances, namely PFOA, PFOS, FOSA, and 6:2 Cl-PFESA, in the brain seemed to be minimal in patients with grade II tumors, as opposed to those with grade IV tumors (Table 1). However, this observation might be coincidental. A vitro cytological study demonstrated that perfluoroalkyl acids induced developmental neurotoxicant actions and the adverse effects of PFAS on neurodevelopmental followed the sequence of PFOSA > PFOS > PFBS ≈ PFOA (Slotkin et al., 2008). Perfluorooctane sulfonamide triggered the highest levels of oxidative stress and cell loss in both undifferentiated and differentiating cells, and even altered the differentiation fate of the cells (Slotkin et al., 2008). Furthermore, FOSA was considered to have a

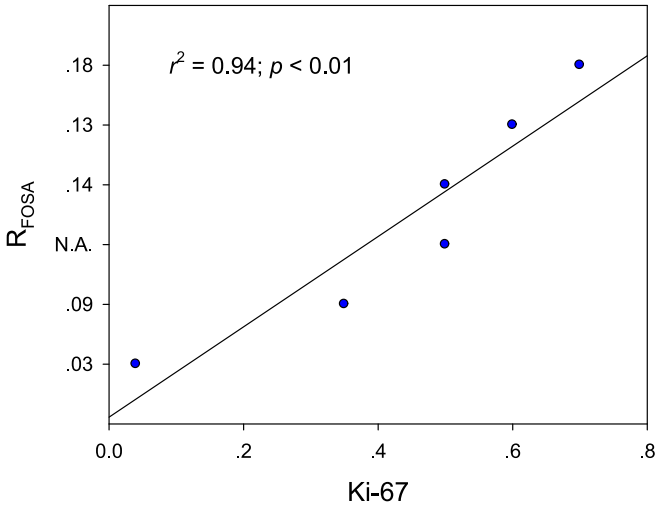

Fig. 4. Linear regression relationship between the blood–brain barrier transmission efficiency of perfluorooctane sulfonamide (FOSA), R_{FOSA}, defined as the concentration ratio in paired glioma tissue and plasma samples, and glioma medical factor Ki-67.

stronger ability than dieldrin and chlorpyrifos to induce cell loss in terms of higher rank for lipid peroxidation (Slotkin and Seidler, 2010). These findings suggest that FOSA exposure could induce potential adverse effects on neurological disorders including glioma. However, our current understanding of the underlying mechanisms for the tumorigenic hazards of FOSA in brain tumors and the factors affecting its penetration of the BBB is still limited. Further studies are needed to explore and elucidate the signaling pathway mechanisms related to FOSA and its role in the occurrence or development of brain tumors. Naturally, it is imperative to acknowledge certain limitations inherent in the present pilot study, including the limited sample number, the inclusion of samples exclusively from patients with brain tumors, and the absence of control samples due to the unique nature of brain tissue samples. These factors may potentially impact the analysis and interpretation of the results, such as statistical random errors due to insufficient sample number, the inadequacy of brain tumor samples alone to account for blood–brain transport efficiencies under non-disease influences, and the specificity of brain tissue samples that have BBB, which makes it impossible to apply the findings to the assessment and prediction of transport efficiencies in other organs.

4. Conclusions

To the best of our knowledge, this is the first study revealing the BBB transmission and accumulation efficiency of PFAS through an examination of the concentration ratios of PFAS in brain tissue and paired plasma samples. Both short- and long-chain PFAS could cross the BBB, but they have low accumulation efficiency in human brain tissue. Among low molecular weight PFAS (including short-chain PFAS and emerging PFAS), 6:2 Cl-PFESA may not be a suitable substitute for PFOS and more attention about its potential ecological health risks shall be paid in future studies. In addition, FOSA, one of the high molecular weight PFAS (long-chain PFAS, including long-chain PFCAs, long-chain PFSAs, and precursor), has been shown to have an association with the occurrence or development of glioma. It should be recognized that the conclusions drawn from the present study should be considered preliminary due to the difficulty in obtaining a large number of paired brain issue and plasma samples. Besides, the interferences of some confounding factors, such as possible degradation of precursor compounds, occurrence of multiple exogenous pollutants, and altered physiological conditions in brain tumor progression, on the BBB were not taken into account. Therefore, the assessment of PFAS transport efficiency in plasma-brain tissue in the present pilot study was exploratory in nature. Future studies based on more paired samples and influencing factors are needed to better characterize human brain exposure to environmental chemicals.

CRediT authorship contribution statement

Meng-Yi Xie: Methodology, Investigation, Conceptualization. **Zhi-Ying Lin:** Resources, Investigation. **Xiang-Fei Sun:** Validation, Methodology. **Jing-Jing Feng:** Validation, Investigation. **Lei Mai:** Supervision, Funding acquisition. **Chen-Chou Wu:** Validation, Supervision, Conceptualization. **Guang-Long Huang:** Resources. **Po Wang:** Validation, Methodology. **Ya-Wei Liu:** Resources. **Liang-Ying Liu:** Validation, Supervision, Funding acquisition. **Eddy Y. Zeng:** Writing – review & editing, Supervision, Project administration, Funding acquisition.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envint.2024.108719.

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